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GUDRUN E. HUCKETT DRAUDT
SCHUBERTSTR. 15A
WUPPERTAL, 42289
GERMANY

EXAMINER	
THOMAS, DAVID C	

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1637	

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Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	10/562,441	BURGGRAF, SIEGFRIED	
	Examiner	Art Unit	
	David C. Thomas	1637	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *27 June 2007*.

2a)☒ This action is **FINAL**. 2b)☐ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *25-32 and 34-47* is/are pending in the application.

4a) Of the above claim(s) *45-47* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *25-32 and 34-44* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____.
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

DETAILED ACTION

1. Applicant's amendment filed June 27, 2007 is acknowledged. Claims 25, 28-30, 34, 35, 38 and 40-43 (currently amended) and 26, 27, 31, 32, 36, 37, 39 and 44 (previously presented) will be examined on the merits. Claims 45-47 were previously withdrawn. Claim 33 has been canceled.

Claim Rejections - 35 USC § 103

2. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

3. Claims 25-32, 34-39 and 42 are rejected under 35 U.S.C. 103(a) as being unpatentable over Wittwer et al. (U.S. Patent No. 6,174,670) in view of Pasloske et al. (U.S. Patent No. 6,399,307).

Wittwer teaches a method for qualitative or quantitative detection of a nucleic acid in a sample (for overview, see Abstract and column 4, lines 32-50), said method comprising the steps of:

amplifying a nucleic acid to be detected in a sample in the presence of at least one single-stranded detection probe that by a reversible binding action binds reversibly to a binding region of said nucleic acid to be detected and enables a detection of said nucleic acid to be detected based on said reversible binding action (target DNA is amplified using pair of primers in presence of two probes which hybridize to adjacent regions of target during annealing phase of PCR at each cycle, column 3, 58-61,

column 7, lines 56-67 and Figure 18, top example; see Figure 2 for probe annealing during phase of lowered temperature);

providing a single-stranded control nucleic acid in said sample and amplifying said single-stranded control nucleic acid in said sample, wherein said single-stranded control nucleic acid has a binding region that also binds said at least one single-stranded detection probe and wherein said binding region of said single stranded control nucleic acid has a nucleotide sequence having at least one deviation in comparison to said nucleotide sequence of said binding region of said nucleic acid to be detected (two nucleic acid targets having difference at selected locus are present in reaction which have binding sites for two probes which hybridize at adjacent sites in target wherein one of the probes spans the locus site and is matched with one of the targets and therefore deviates from the sequence of control target, column 7, lines 53-54 and line 64 to column 8, line 9; see Figure 18 for dual probe annealing; genomic DNA that is provided as the control DNA is denatured during the initial amplification step, and therefore is in single-stranded form, while the nucleic acid of some pathogens such as HIV used for the testing of viral load in patients infected with HIV for purposes of prognosis and therapy is inherently single-stranded (column 11, lines 54-60).

wherein a first product of said nucleic acid to be detected and of said at least one single-stranded detection probe and a second product of said single-stranded control nucleic acid and of said at least one single-stranded detection probe have different melting points and a temperature difference of said melting points is sufficiently large to analytically differentiate said first and second products from one another for carrying out

said detection (temperature melting profiles for each target sample are generated that are distinguishable if a true sequence deviation exists between the targets, column 7, line 64 to column 8, line 9 and column 8, lines 25-35; see also Example 23, column 46, lines 20-48, for detection of heterozygous and homozygous forms of methylenetetrahydrafolate reductase (MTHFR) gene and Figure 48), wherein said detection is carried out at a temperature that is 2°C to 10°C below said melting temperature of said first product (monitoring of fluorescence begins at 50°C during melting profile measurements, more than 2°C but less than 10°C below melting temperature of homozygous mutant control for MTHFR gene example, column 46, lines 42-46 and Figure 48).

With regard to claim 26 and 27, Wittwer teaches a method for qualitative or quantitative detection of a nucleic acid in a sample wherein said melting point of said second product is lower than said melting point of said first product by at least 5°C. (for analysis of point mutation in MTHFR gene, melting point of homozygous mutant is lower than that of wild-type by about 5°C, column 46, lines 20-48 and Figure 48).

With regard to claim 28, Wittwer teaches a method for qualitative or quantitative detection of a nucleic acid in a sample wherein said single-stranded control nucleic acid and said nucleic acid to be detected are amplified with identical primers (amplification of both control and target nucleic acids are performed with same primers, column 46, lines 27-34).

With regard to claim 29, Wittwer teaches a method for qualitative or quantitative detection of a nucleic acid in a sample wherein said nucleic acid to be detected and said

single-stranded control nucleic acid are amplified by polymerase chain reaction (amplification of control and target nucleic acids is achieved by PCR, column 8, lines 10-18 and column 46, lines 34-46).

With regard to claim 30, Wittwer teaches a method for qualitative or quantitative detection of a nucleic acid in a sample wherein two or more of said nucleic acid to be detected and two or more of said single-stranded control nucleic acid are present in said same sample and wherein for each one of said nucleic acids to be detected one of said single-stranded control nucleic acids is present (the discriminatory power of hybridization probes can be applied to multiplex PCR using multiple detection probes that sequentially melt off different targets at different temperatures, column 46, lines 49-61).

With regard to claim 31, Wittwer teaches a method for qualitative or quantitative detection of a nucleic acid in a sample wherein said nucleic acid to be detected is a DNA or an RNA derived in particular from a pathogen (nucleic acid from pathogens such as hepatitis B and C, and HIV can be detected using hybridization probes that distinguish wild-type and variants by melting curve profiles, column 35, lines 30-60 and column 41, lines 54-60).

With regard to claim 32, Wittwer teaches a method for qualitative or quantitative detection of a nucleic acid in a sample wherein said detection of said nucleic acid to be detected is carried out in real-time (monitoring of factor V Leiden mutation can be monitored both in real time during each cycle, as well as by performing melting profile

after completion of amplification, column 44, line to column 45, line 14 and Figures 46 and 47).

With regard to claim 34, Wittwer teaches a method for qualitative or quantitative detection of a nucleic acid in a sample wherein said melting point of said second product is so low that said second product is negligible or not at all present in said detection (melting point of homozygous mutant is comparatively low for factor V Leiden mutation and product appears not be present when monitoring sample at 63°C where both heterozygous and homozygous wild-type are not fully melted, column 44, line 65 to column 45, line 14 and Figure 47).

With regard to claim 35, Wittwer teaches a method for qualitative or quantitative detection of a nucleic acid in a sample wherein only one of said at least one single-stranded detection probe is used and said detection of said nucleic acid to be detected is based on a melting curve of said nucleic acid to be detected in the presence of said at least one single-stranded detection probe, wherein a melting curve of said single-stranded control nucleic acid in the presence of said at least one single-stranded detection probe serves as an internal control of proper amplification (example of melting profile for monitoring MTHFR gene mutation is performed using one labeled probe along with a labeled primer, wherein control nucleic acid is amplified and monitored by performing melting curve, which is indicative of proper amplification, column 46, lines 20-48 and Figure 48).

With regard to claim 36, Wittwer teaches a method for qualitative or quantitative detection of a nucleic acid in a sample wherein two of said at least one single-stranded

detection probe are used, wherein a first one of said two single-stranded detection probes carries a reporter group and a second one of said two single-stranded detection probes changes observable properties of said reporter group when in a position in the vicinity of said reporter group (two labeled hybridization probes can be used wherein the probes hybridize at closely spaced sites on the target, with one probe being 3'-labeled with fluorscein to allow transfer of energy to nearby Cy5 reporter group on 5' end of second probe, column 31, line 43 to column 32, line 7 and Figure 18).

With regard to claim 37, Wittwer teaches a method for qualitative or quantitative detection of a nucleic acid in a sample wherein said at least one single-stranded detection probe carries a reporter group and a second group that changes observable properties of said reporter group when in a position in the vicinity of said reporter group, wherein said reporter group and said second group are positioned so close to one another that said observable properties of said reporter group are changed either only during binding of said at least one single-stranded detection probe to said nucleic acid to be detected or only in a non-bonded state of said at least one single-stranded detection probe (dual-labeled probes can be used wherein the reporter and second group are subject to quenching of the fluorescence signal when not hybridized, wherein observable properties are changed upon binding to a target during amplification whereupon the probe is cleaved to further increase the signal, column 21, lines 47-49, column 28, lines 48-55, and Figure 5B).

With regard to claim 38, Wittwer teaches a method for qualitative or quantitative detection of a nucleic acid in a sample wherein said nucleotide sequence of said single-

stranded control nucleic acid in said binding region for said at least one single-stranded detection probe has at least one modification relative to said nucleic acid to be detected (single base mutations are detected in samples containing factor V Leiden mutation (column 42, lines 62-64) as well as MTHFR gene mutation, column 46, lines 20-26).

With regard to claim 39, Wittwer teaches a method for qualitative or quantitative detection of a nucleic acid in a sample wherein said at least one modification is an exchange of a G or a C (factor V Leiden mutation involves G to A mutation, column 42, lines 62-64, while MTHFR gene mutations involves C to T mutation, column 46, lines 20-26).

With regard to claim 42, Wittwer teaches a method for qualitative or quantitative detection of a nucleic acid in a sample wherein a sequence region of said single-stranded control nucleic acid that can neither hybridize with said at least one single-stranded detection probe nor optionally with a primer is shortened (probes can be designed to detect deletion mutations, such that a shortened version of the control nucleic acid will have lower melting temperature and thus not hybridize at certain temperatures during melting profile, column 45, lines 60-67).

Wittwer does not teach a method for qualitative or quantitative detection of a nucleic acid in a sample comprising the step of adding a single-stranded control nucleic acid to the sample.

Pasloske teaches a method of quantifying viral load in an animal using a ribonuclease-resistant RNA preparation that is added as an internal control to reverse transcription and PCR amplification steps during viral load measurements such as for

HIV, HCV and HTLV (column 5, lines 39-47 and column 6, lines 1-11). Pasloske also teaches internal control RNA molecules that are differentiated from wild-type molecules by having altered sequences between the common primer binding sites such as incorporation of a restriction site or a deletion (column 30, lines 1-12).

Pasloske does not teach a method for qualitative or quantitative detection of a nucleic acid in a sample comprising detection of target and control nucleic acids in the presence of a single-stranded probe wherein the binding regions of the target and control nucleic acid sequences for the single-stranded probe deviate by at least one nucleotide. Pasloske also does not teach a method wherein the amplification products of the target and control nucleic acids have different melting points to analytically differentiate the products, wherein the detection is carried out at a temperature that is 2°C to 10°C below said melting temperature of the product of the target nucleic acid.

It would have been prima facie obvious to one of ordinary skill in the art at the time the invention was made to combine the methods of Wittwer for monitoring and detecting nucleic acid hybridization of probes to target and control molecules using melting curve analysis and those of Pasloske who teaches methods of viral load measurement by addition of single-stranded nucleic acid controls to target samples since the quantitative methods of Wittwer are also useful for measurement of relative copy numbers required for quantifying viral loads in patients infected with RNA-based agents such as HIV or hepatitis C (Wittwer, column 41, lines 52-56) that require single-stranded nucleic acid controls. Thus, an ordinary practitioner would have been motivated to use the melting profile methods of Wittwer to analyze target and control

nucleic acids that contain binding regions for detection probes that deviate by one or more nucleotides to allow melting point analysis to differentiate the amplification products during quantification. With regard to viral load measurements, Wittwer states "Using a control template and monitoring the efficiency of amplification of both control and natural templates during amplification, accurate quantification of initial copy number is achieved", column 41, lines 56-60). Further in regard to mRNA quantitation using melting point analysis, Wittwer states "Relative quantification of two PCR products is important in many quantitative PCR applications. Multiplex amplification of two or more products followed by intergration of the areas under melting peaks will be extremely useful in these areas. mRNA is often quantified relative to the amount of mRNA of a housekeeping gene" (Wittwer, column 42, lines 20-25). Finally, the use of an RNA molecule as an internal control for RT-PCR is necessary to control for both the reverse transcription and PCR steps, and the internal standard is generally regarded as more accurate for quantification relative to external standards (Pasloske, column 5, lines 44-47 and column 6, lines 21-23).

4. Claims 40, 41, 43, and 44 are rejected under 35 U.S.C. 103(a) as being unpatentable over Wittwer et al. (U.S. Patent No. 6,174,670) in view of Pasloske (U.S. Patent No. 6,399,307) and further in view of Picard et al. (U.S. Patent No. 6,265,170).

Wittwer and Pasloske together teach the limitations of claims 25-32, 34-39 and 42 as discussed above.

Neither Wittmer nor Pasloske teach a method for qualitative or quantitative detection of a nucleic acid in a sample wherein said nucleotide sequence of said single-stranded control nucleic acid in said binding region for said at least one single-stranded detection probe has at least three modifications relative to said nucleic acid to be detected. Furthermore, neither Wittmer nor Pasloske teach a method wherein the probe fails to hybridize with a probe or primer because of modifications relative to the nucleic acid to be detected, and wherein the modifications are distributed approximately uniformly across said binding region for said at least one single-stranded detection probe.

Picard teaches a method for measuring binding affinity of nucleic acid probes for reference and target molecules by application of voltage to test samples, including probes that contain mismatches or deletions relative to the target such as one- to three-base mismatches and one- to three-base deletions (column 2, line 51 to column 3, line 6) and probes containing 3 noncontiguous mismatches distributed evenly over the probe/target binding region (column 15, lines 17-28).

It would have been prima facie obvious to one of ordinary skill in the art at the time the invention was made to combine the methods of Wittwer and Pasloske for monitoring and detecting nucleic acid hybridization of probes to target and single-stranded control nucleic acid molecules using melting curve analysis and those of Picard for measuring binding affinities of probes containing unusual mutations such as two- and three-base mismatches or deletions, or multiple base mismatches separated by normal base pairs, since the methods of Wittwer are easily capable of measuring any

known mutation including insertions and deletions where a probe can be designed to differ in melting temperature when hybridized to mutant versus wild-type sequences (Wittwer, column 45, lines 60-67). Thus, an ordinary practitioner would have been motivated to use the melting profile methods of Wittwer to analyze the probe/target pairs taught by Picard containing modifications greater than single-base mismatches since probes with these types of changes can be prepared just as easily as those containing single nucleotide changes. Furthermore, since single mismatches impart at least a 4°C shift in the T_m of hybridization probes (Wittwer, column 42, lines 55-59), probes containing additional modifications will be even more readily distinguishable in melting profile analysis.

Response to Arguments

5. Applicant's arguments filed June 27, 2007 have been fully considered but they are not persuasive.

Applicant argues that the rejection of claims 25-39 and 42 under 35 U.S.C. § 102(b) as being anticipated by Wittwer et al. (U.S. Patent No. 6,174,670) should be withdrawn since the reference no longer teaches all the limitations of the claims as amended. In particular, Applicant argues that Wittwer does not disclose that a single-stranded control nucleic acid is added to a sample prior to quantitative detection. The Examiner agrees that Wittwer teaches providing a single-stranded control nucleic acid but does not teach the addition of such a control to a sample. Therefore, 102(b) rejection of claims 25-39 and 42 is withdrawn.

However, upon further searching, another reference was found that teaches the addition of single-stranded control nucleic acids in the form of RNA to RT-PCR assays for quantitative measurements of viral load (Pasloske, column 5, lines 39-47 and column 6, lines 1-11). It is obvious to combine the melting curve analysis methods of Wittwer with the viral load measurement methods of Pasloske since the methods of Wittwer are readily adaptable to assays requiring single-stranded control nucleic acids such as RNAs (Wittwer, column 41, lines 52-56 and column 42, lines 20-25).

Applicant then argues that Wittwer does not disclose a detection method carried out at a temperature that is 2°C to 10°C below the melting temperature of the product of the target nucleic acid since the cited Example and figure in the reference represents comparison of three genotypes, none of which represent a control nucleic acid. The Examiner asserts that Example 23 and Figure 48 represent measurement of melting peaks for related nucleic acid samples, any of which can be considered as a control for the others. Therefore, in this case, it is arbitrary as to what represents a first product and a control. As seen in Figure 48, the melting profile analysis for the homozygous mutant begins at 50°C, which is more than 2°C but less than 10°C below the melting temperature of this nucleic acid. It would be obvious to one or ordinary skill in the art to initiate melting temperature analysis below the melting point of the nucleic acid with the lowest melting point in a mixture of target and control templates in order to fully differentiate between two nucleic acid species in a sample, such as target and single-stranded control nucleic acids. Since Pasloske teaches the addition of single-stranded nucleic acids to such assays, as discussed above, claims 25-32, 34-39 and 42 are now

rejected under 35 U.S.C. § 103(a) as being unpatentable over Wittwer in view of Pasloske.

Finally, Applicant argues that the rejection of claims 40, 41, 43 and 44 under 35 U.S.C. § 103(a) as being unpatentable over Wittwer in view of Picard et al. (U.S. Patent No. 6,265,170) should be withdrawn since the reference no longer teaches all the limitations of the claims as amended. This rejection is not separately argued on the merits and therefore the 103 rejection, with the addition of Pasloske, is maintained since the base claim has been determined to be unpatentable over Wittwer and Pasloske, as discussed above.

Summary

6. Claims 25-32 and 34-44 are rejected. No claims are allowable.

Conclusion

7. Applicant's amendment necessitated the new ground(s) of rejection presented in this Office action. Accordingly, **THIS ACTION IS MADE FINAL**. See MPEP § 706.07(a). Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of

the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the date of this final action.

Correspondence

8. Any inquiry concerning this communication or earlier communications from the examiner should be directed to David C. Thomas whose telephone number is 571-272-3320 and whose fax number is 571-273-3320. The examiner can normally be reached on 5 days, 9-5:30.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Gary Benzion can be reached on 571-272-0782. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

David C. Thomas 9/11/07

David C. Thomas
Patent Examiner
Art Unit 1637

JEFFREY FREDMAN
PRIMARY EXAMINER
9/18/07